Downregulation of mouse intestinal Na(+)-coupled glucose transporter SGLT1 by gum arabic (Acacia Senegal)

Nasir, O ; Artunc, F ; Wang, K ; Rexhepaj, R ; Föller, M ; Ebrahim, A ; Kempe, D S ; Biswas, R ; Bhandaru, M ; Walter, M ; Mohebbi, N ; Wagner, Carsten A ; Saeed, A M ; Lang, F

Abstract: Intestinal Na(+)-coupled glucose transporter SGLT1 determines the rate of glucose transport, which in turn influences glucose-induced insulin release and development of obesity. The present study explored effects of Gum Arabic (GA), a dietary polysaccharide from dried exudates of Acacia Senegal, on intestinal glucose transport and body weight in wild-type C57Bl/6 mice. Treatment with GA (100 g/l) in drinking water for four weeks did not affect intestinal SGLT1 transcript levels but decreased SGLT1 protein abundance in jejunal brush border membrane vesicles. Glucose-induced jejunal short-circuit currents revealed that GA treatment decreased electrogenic glucose transport. Drinking a 20% glucose solution for four weeks significantly increased body weight and fasting plasma glucose concentrations, effects significantly blunted by simultaneous treatment with GA. GA further significantly blunted the increase in body weight, fasting plasma glucose and fasting insulin concentrations during high fat diet. In conclusion, the present observations disclose a completely novel effect of gum arabic, i.e. its ability to decrease intestinal SGLT1 expression and activity and thus to counteract glucose-induced obesity.

DOI: https://doi.org/10.1159/000276554

Original Paper

Cellular Physiology and Biochemistry

Cell Physiol Biochem 2010;25:203-210

Accepted: October 08, 2009

Downregulation of Mouse Intestinal Na^+-coupled Glucose Transporter SGLT1 by Gum Arabic (*Acacia Senegal*)

Omaima Nasir[a], Ferruh Artunc[b], Kan Wang[a], Rexhep Rexhepaj[a], Michael Föller[a], Ammar Ebrahim[a], Daniela S. Kempe[a], Raja Biswas[a], Madhuri Bhandaru[a], Michael Walter[c], Nilufar Mohebbi[d], Carsten A. Wagner[d], Amal M. Saeed[e] and Florian Lang[a]

[a]Department of Physiology, University of Tuebingen, [b]Internal Medicine IV, University of Tuebingen, [c]Human Genetics, University of Tuebingen, [d]Department of Physiology, University of Zurich, [e]Department of Physiology, University of Khartoum

Key Words

Gum arabic • Glucose transport • Obesity • Hyperglycemia • Hyperinsulinism

Abstract

Intestinal Na^+-coupled glucose transporter SGLT1 determines the rate of glucose transport, which in turn influences glucose-induced insulin release and development of obesity. The present study explored effects of Gum Arabic (GA), a dietary polysaccharide from dried exudates of *Acacia senegal*, on intestinal glucose transport and body weight in wild-type C57Bl/6 mice. Treatment with GA (100 g/l) in drinking water for four weeks did not affect intestinal SGLT1 transcript levels but decreased SGLT1 protein abundance in jejunal brush border membrane vesicles. Glucose-induced jejunal short-circuit currents revealed that GA treatment decreased electrogenic glucose transport. Drinking a 20% glucose solution for four weeks significantly increased body weight and fasting plasma glucose concentrations, effects significantly blunted by simultaneous treatment with GA. GA further significantly blunted the increase in body weight, fasting plasma glucose and fasting insulin concentrations during high fat diet. In conclusion, the present observations disclose a completely novel effect of gum arabic, i.e. its ability to decrease intestinal SGLT1 expression and activity and thus to counteract glucose-induced obesity.

Introduction

Gum Arabic (GA) is a water-soluble dietary fiber derived from the dried gummy exudates of the stems and branches of *Acacia senegal* [1]. Chemically, GA is a polysaccharide based on branched chains of (1-3) linked β-D-galactopyranosyl units. Side chains of 2-5 units in length are attached by (1-6) links to the main chain. Both, the main chain and the side chains contain α-L-arabinofuranosyl, α-L-rhamnopyranosyl, β-D-glucuronopyranosyl and 4-O-methyl-β-D-glucuronopyranosyl units [2]. GA is readily soluble in water without increasing viscosity [3].

GA is widely used as an emulsifier and stabilizer by the pharmaceutical and the food industry. It is primarily indigestible for both humans and animals, and after passing the small intestine it is fermented under the influence of microorganisms in the colon to short chain fatty acids [4]. The US Food and Drug Administration recognized it as one of the safest dietary fibers [5].

KARGER

Fax +41 61 306 12 34
E-Mail karger@karger.ch
www.karger.com

1015-8987/10/0253-0203$26.00/0

Accessible online at:
www.karger.com/cpb

Florian Lang
Department of Physiology, University of Tuebingen
Gmelinstr. 5, 72076 Tuebingen (Germany)
Tel. +49-7071-2972194, Fax +49-7071-295618
E-Mail florian.lang@uni-tuebingen.de

GA exhibits pharmacological effects related to interference with gastrointestinal absorption of nutrients. In a previous study we could show that GA blunted intestinal absorption of Na^+ and water in healthy mice while enhancing calcium and magnesium uptake [6]. In a rat model of chronic osmotic-diarrhea, GA exerted pro-absorptive properties by increased sodium and water absorption [7-9]. Thus, we hypothesized that GA treatment may also influence the intestinal glucose absorption, which is mainly accomplished by the Na^+-coupled glucose transporter SGLT1 [10]. To test this hypothesis, we utilized gene array analysis and/or western blotting to quantify the expression of SGLT1 as well as known modulators of intestinal Na^+-coupled glucose transport such as RS1 (RSC1A1) [11], serum- and glucocorticoid-inducible kinase isoforms SGK1 and SGK3 [12] in small intestine from wild-type C57Bl/6 mice. We further determined electrogenic glucose transport in small intestine and investigated, whether GA treatment influences hyperglycemia, hyperinsulinemia and body weight gain following ingestion of glucose-rich or high fat diet.

Materials and Methods

Gum Arabic

GA in powder form was a generous gift from Dar Savanna Ltd., Khartoum, Sudan. It is a 100% natural extract powder produced mechanically from the wildly grown *Acacia senegal* tree with a particle size less than 210 µm. The quality conformed to the food and pharmaceutical requirements of the Food and Agriculture Organization of the United Nations (FAO), British pharmacopoiea (BP), United States pharmacopoiea (USP) and Joint FAO/WHO Expert Committee On Food Additives (JECFA).

Animals

Experiments were carried out in male 8-12 week old wild-type C57Bl/6 mice (Charles River, Germany). The animals were housed under controlled environmental conditions (22-24°C, 50-70% humidity and a 12-h light/dark cycle). Throughout the study mice had free access to standard pelleted food containing 4.5% crude fiber (C1310, Altromin, Lage, Germany) and tap water or Gum Arabic solution as indicated. All animal experiments were conducted according to the German law for the care and welfare of animals and had been approved by local authorities.

Animal experimentation

Animals were provided with a control diet (C1310, 4 kcal% fat, 0.25% Na^+, 0.36% Cl^-, 0.71% K^+, Altromin, Heidenau, Germany) or a high fat diet (C1000, 45 kcal% fat, 0.25% Na^+, 0.36% Cl^-, 0.71% K^+, modified according D12451 from Research Diet, Altromin, Heidenau, Germany) and received 10% (w/v) GA dissolved in tap water (100 g/l) as indicated; preparations were refreshed every 3 days during the treatment.

Glucose and insulin concentrations were determined in blood drawn after tail-vein bleeding. Plasma glucose was determined using a glucometer after overnight fasting (Accutrend, Roche, Mannheim, Germany); plasma insulin was measured using an ELISA (Crystal Chem INC, USA).

To investigate the effects of GA treatment on the development of hyperglycemia, hyperinsulinemia and obesity following excessive glucose intake, mice were housed individually and 20% (w/v) glucose added to the drinking water either with or without 10% GA. Control groups received tap water or 10% GA. During the treatment for 4 weeks, body weight, food and fluid intake were monitored.

Gene array analysis

To investigate the effect of GA treatment on intestinal gene expression, whole genome gene expression profiling was performed in intestinal tissue of untreated mice and of mice treated with GA for one week (each n = 5). 1 µg of total RNA was linearly amplified and biotinylated using the One-Cycle Target Labeling Kit (Affymetrix, Santa Clara, CA, USA) according to the manufacturer's instructions. 15 µg of labeled and fragmented cRNA was hybridized to MOE 430 2.0 mouse whole genome Gene Chip® arrays (Affymetrix). After hybridization the arrays were washed and stained in a Fluidics Station 450 (Affymetrix) with the recommended washing procedure. Biotinylated cRNA bound to target molecules was detected with streptavidin-coupled phycoerithrin, biotinylated anti-streptavidin IgG antibodies and again streptavidin-coupled phycoerithrin according to the protocol. Arrays were scanned using the GCS3000 Gene Chip scanner (Affymetrix) and GCOS 1.4 software. Scanned images were subjected to visual inspection to control for hybridization artifacts and proper grid alignment and analyzed with Microarray Suite 5.0 (Affymetrix) to generate report files for quality control.

For statistical data analysis the CEL-files were imported into Genespring 7.1 (Agilent Technologies, Santa Clara, CA) using Genespring's implementation of GC-RMA for normalization and probe summarization. For all transcripts the average change in expression (fold change) and the p-value using Welch's t-test was calculated. The effect of GA treatment on the expression of selected transcripts was examined.

Immunoblotting of SGLT1 from small intestine

The intestinal membrane expression of SGLT1 was determined in brush-border membrane vesicles (BBMV) prepared from untreated mice, mice treated with GA for four weeks, mice treated with 20% glucose for four weeks and mice treated with both 10% GA and 20% glucose for four weeks using the Mg^{2+} precipitation technique [13]. Briefly, mice were killed with CO_2 and the whole intestine was removed and divided into two parts. After discarding the first 2 cm as duodenum the next 13 cm were considered as jejunum and the remainder as ileum. Jejunum and ileum were opened longitudinally. Contents were washed out with ice-cold PBS. The epithelial layer was scraped off with a glass slide in a buffer containing (in mM) 250 sucrose, 20 Tris (pH 7.5), 5 EGTA and a protease inhibitor

Table 1. Gene array analysis of major genes involved in intestinal glucose uptake. Relative change in expression of the respective transcript after one week of GA treatment (n= 5 chips each).

Represented gene	Name of encoded protein	Systematic transcript name	Fold change	p-value
slc5a9	SGLT1	1426634_at	-1.05	*0.492*
		1452136_at	1.07	*0.074*
		1439494_at	-1.03	*0.483*
		1440779_s_at	1.05	*0.634*
rsc1a1	RSC1	1435536_at	1.25	*0.008*
sgk1	SGK1	1416041_at	1.93	*0.006*
sgk3	SGK3	1450036_at	-1.13	*0.003*
slc2a2	GLUT2	1449067_at	1.18	*0.071*

cocktail (Roche, Mannheim, Germany). The suspension was homogenized with a Sorvall Omnimixer (rotating blade). $MgCl_2$ was added to the homogenate yielding a final concentration of 10 mM. The suspension was stirred on ice and then centrifuged at 1,600 *g* for 15 min. The plasma membranes retained in the supernatant were collected by centrifugation at 20,000 *g* for 30 min. The resultant pellet was suspended in a pH 7.4 buffer consisting of (in mM) 125 sucrose, 10 Tris (pH 7.5), 2.5 EGTA, 2.5 $MgSO_4$. This suspension was homogenized with 50 up-down strokes using a glass homogenizer and centrifuged at 20,000 *g* for 30 min. The final pellet containing the purified BBMV was homogenized by passing the suspension through 25- and 28-gauge needles and solubilised. All steps were carried out at 4°C. Samples from three animals per group were pooled and frozen for later use. Western blotting was performed in three to four independent experiments, resulting in a total of n = 9-12 mice in each group.

After measurement of the total protein concentration (Biorad Protein kit), 30 µg of brush border membrane protein were solubilized in Laemmli sample buffer at 95°C for 5 min, and SDS-Page was performed on 10% polyacrylamide gels. For immunoblotting, proteins were transferred electrophoretically from unstained gels to polyvinylidene difluoride membranes (Immobilon-P, Millipore, Bedford, MA, USA). After blocking with 5% milk powder in Tris-buffered saline/0.1% Tween-20 for 60 min, the blots were incubated with affinity purified rabbit anti-mouse SGLT1 antibody (1:2000; 55 kDa, Chemicon, Temecula, CA, USA) overnight at 4°C. After washing blots were incubated with secondary antibodies linked with horse radish peroxidase (goat anti-rabbit 1: 2000 Cell Signaling, USA) for 1 h at room temperature. Antibody-binding was detected with the ECL Western blotting detection kit (RPN 2106 GE health care, UK). Bands were quantified using Quantity One Software (Biorad, München, Germany).

To identify the specificity of the band, the control peptide was applied. The diluted control peptide (1:1000, AG661 Chemicon, US) was mixed with the SGLT1 antibody for 30 min at room temperature before incubation with the other blot, which was done in parallel.

To provide the loading control the blot was stripped in stripping buffer (Roth, Karlsruhe, Germany) at 56°C for 30 min. After washing with TBST, the blot was blocked with TBST + 5% milk for 1 h at room temperature, then incubated with rabbit anti-mouse β-actin antibody (1:1000, Cell Signaling) at 4°C over night. After washing the blots were incubated with secondary antibodies linked with horse radish peroxidase (goat anti-rabbit 1: 2000, Cell Signaling), for 1 h at room temperature. Antibody-binding was detected with the ECL Western blotting detection kit (RPN 2106 GE health care, UK). Bands were quantified with Quantity One Software (Biorad, München, Germany).

Ussing chamber experiments in small intestine

For the analysis of electrogenic intestinal glucose transport, Ussing chamber experiments were performed in mice after 1 week treatment with or without GA. Following treatment mice were killed by cervical dislocation and the intestine was quickly removed. Jejunal segments (5-10 cm postpylorus) were mounted into a custom-made mini-Ussing chamber with an opening diameter of 0.99 mm and an opening area of 0.00769 cm^2. Under control conditions, the serosal and luminal perfusate contained (in mM) 105 NaCl, 2 KCl, 1 $MgCl_2$, 1.25 $CaCl_2$, 0.4 KH_2PO_4, 1.6 K_2HPO_4, 5 Na pyruvate, 25 $NaHCO_3$, and 20 mannitol. Where indicated, 10 or 20 glucose, 20 L-glutamine, 20 L-phenylalanine, 20 L-proline or 20 L-methionine were added to the luminal perfusate at the expense of mannitol. All solutions were gassed with 95% O_2-5% CO_2 for at least 60 min until usage in the experiment. GA was not present in any of the substrate solutions of GA-treated mice. All substances were from Sigma (Schnelldorf, Germany) or Roth (Karlsruhe, Germany).

Statistics

Data are provided as means ± SEM, *n* represents the number of independent experiments. All data were tested for significance with one-way ANOVA followed by Tukey-Kramer post-test, or unpaired Student's t-test, as appropriate using GraphPad InStat version 3.00 for Windows 95, GraphPad Software, San Diego, California, USA. A p-value < 0.05 was considered statistically significant.

Results

To determine, whether treatment with GA influences the transcript levels of SGLT1 and/or regulators of the carrier, gene array analysis was performed prior to and following GA treatment for one week. As listed in table 1, GA treatment did not significantly modify SGLT1 transcript levels. The transcript levels of SGK1, a known stimulator of SGLT1 [14], were even significantly increased. However, GA treatment significantly decreased the transcript levels of SGK3, another known stimulator of SGLT1 [14] and increased the transcript

Fig. 1. Effect of GA on SGLT1 protein abundance in brush-border membrane vesicles from jejunum and ileum. Original western blots showing SGLT1 and actin protein abundance in preparations of jejunum (A) and ileum (B) from untreated mice, mice treated with either gum arabic (GA) or glucose (glc) or both for four weeks. The right panels depict the arithmetic means ± SEM (n = 3-4) of the relative intensity of the SGLT1 protein band. * indicates statistically significant difference ($p<0.05$; paired t-test).

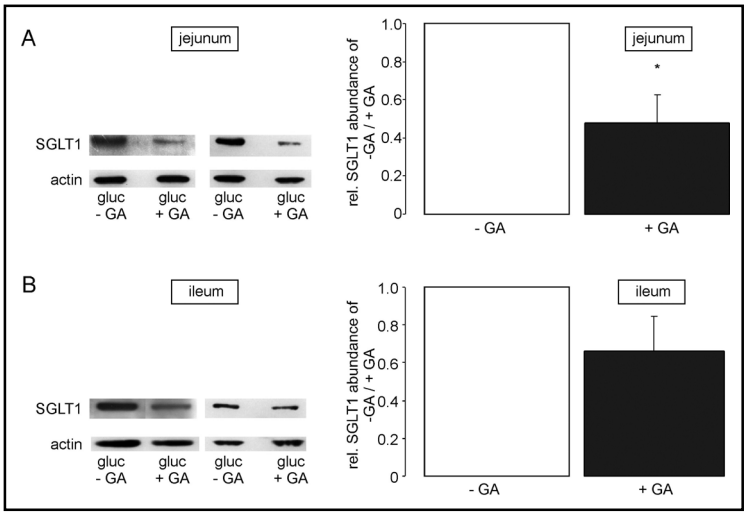
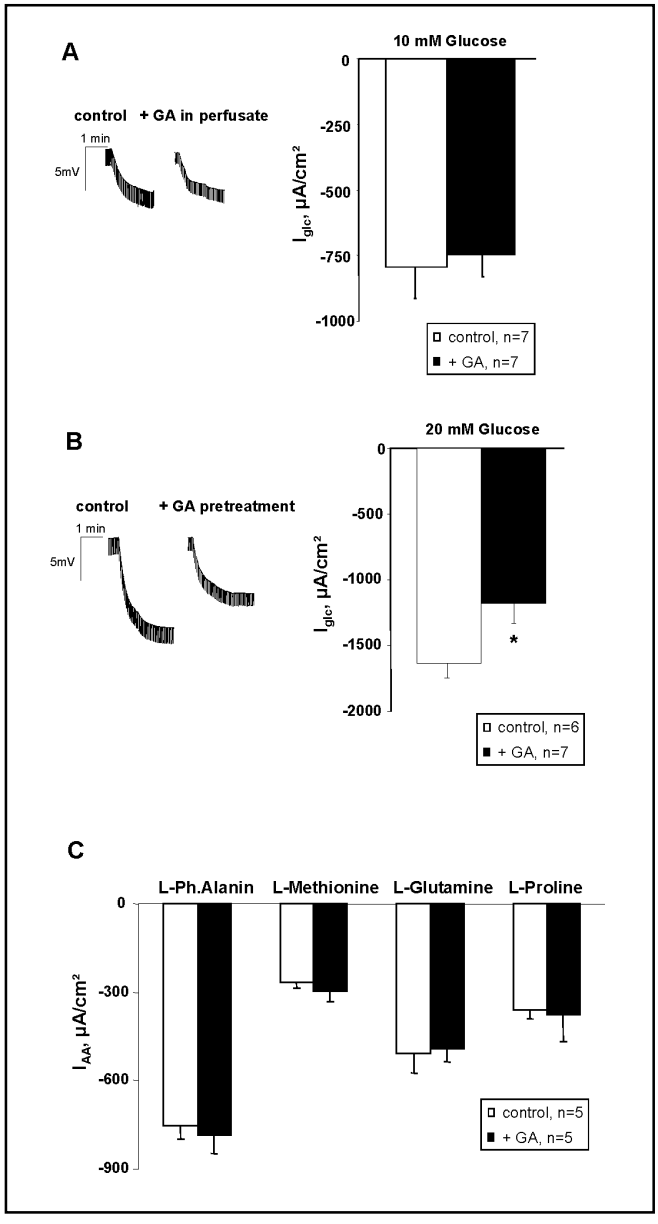


Fig. 2. Effect of GA treatment on intestinal electrogenic substrate transport. A Original tracings (left) and arithmetic means (right) of the effect of GA treatment on glucose-induced short-circuit current (I_{glc}) in untreated mice with (solid bar) or without (open bar) 10% GA in the perfusate. Due to the high viscosity experiments were conducted with 10 mM glucose. B Original tracings (left) and arithmetic means (right) of the effect of GA on glucose-induced short-circuit current (I_{glc}) in untreated mice (open bar) and mice treated with GA for one week (solid bar). Note that in those experiments GA was not added to the perfusate. C Arithmetic means ± SEM of amino acid-induced short circuit currents (I_{AA}) in proximal jejunum from untreated mice (open bars) and GA-treated mice (solid bars; unpaired t-test, $p<0.05$). *indicates significant difference between control and GA treatment.



levels of RSC1, a known transcriptional and posttranscriptional inhibitor of SGLT1 [11]. GLUT2 mRNA expression was not changed significantly under GA treatment (Table 1).

In a second step, Western blotting was utilized to clarify, whether GA treatment influences SGLT1 protein expression (Fig. 1A). Immunoblotting of brush-border membranes indeed disclosed significantly decreased SGLT1 protein abundance in jejunum. In ileum, GA treatment tended to similarly decrease SGLT1 expression, an effect, however, not reaching statistical significance. (Fig. 1B).

The influence of GA treatment on SGLT1 protein abundance in jenunal brush border membranes prompted us to study the interaction of GA with the intestinal absorption of glucose by determining electrogenic glucose transport in jejunal segments. In untreated mice, the transepithelial potential difference ($V_{t,p}$) and the transepithelial resistance ($R_{t,p}$) amounted to 1.08 ± 0.06

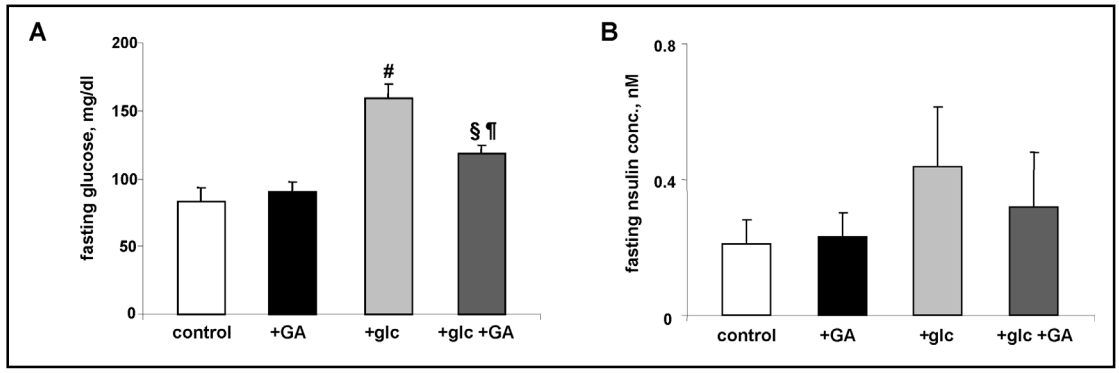


Fig. 3. Effect of GA treatment on fasting blood glucose and plasma insulin concentrations. A & B: Arithmetic means ± SEM of fasting blood glucose and plasma insulin concentrations in mice treated with tap water (control, n = 10), GA (n = 12), 20% glucose and glucose + GA (n = 20 each). [#] indicates significant difference between control and glucose treatment, [¶]indicates significant difference between GA and glucose + GA treatment, [§]indicates significant difference between glucose and glucose + GA treatment.

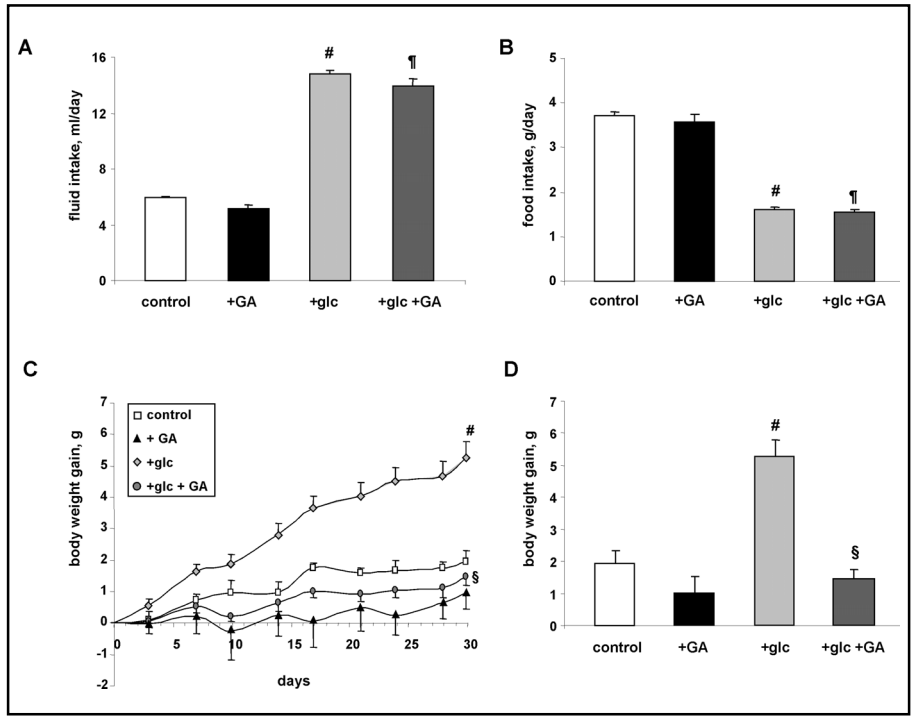


Fig. 4. Effects of GA treatment on glucose-induced obesity. A & B: Arithmetic means ± SEM of mean daily fluid and food intake in untreated mice (n = 11) and mice treated with either GA (n = 12) or glucose (n = 20) or both glucose and GA (n = 20) for four weeks (one way-ANOVA with Tukey´s post-test, $p<0.0001$). C Arithmetic means ± SEM of the body weight gain during a 4 week treatment of mice with tap water, with GA, with 20% glucose or with glucose + GA. D Arithmetic means ± SEM of total body weight gain after a 4 week treatment with GA, with 20% glucose or with glucose + GA. [#]indicates significant difference between control and glucose treatment, [¶]indicates significant difference between GA and glucose+GA treatment, [§]indicates significant difference between glucose and glucose+GA treatment.

mV and 8.33± 0.46 Ω·cm² (n = 7), respectively. The isoosmotic replacement of mannitol by glucose generated a lumen-negative shift of the transmural potential difference without significantly altering the transmural resistance. The glucose-induced alterations of transepithelial voltage and transepithelial resistance allowed the calculation of the glucose-induced short-circuit current I_{glc}. In jejunal segments of untreated mice, I_{glc} amounted to -1636 ± 112 µA/cm² at 20 mM glucose. Addition of 10 % GA to the luminal perfusate of intestinal segments from untreated mice did not significantly modify glucose-induced short-circuit currents, indicating that GA did not directly interfere with luminal glucose or the carrier (Fig. 2A). However, as shown in Fig. 2B, GA treatment for 1 week decreased glucose-induced short-circuit current to -1179 ± 148 µA/cm² without altering $V_{t,p}$ (1.01 ± 0.06 mV, n =6) or $R_{t,p}$ (7.74 ± 0.29 Ω·cm², n = 6). To explore whether GA interferes with other sodium-coupled transport processes, amino acid-induced currents were studied in addition. As shown in Figure 2C, GA treatment did not significantly alter the electrogenic transport of glutamine, proline, phenylalanine and methionine. Hence, GA treatment rather selectively inhibited intestinal glucose absorption.

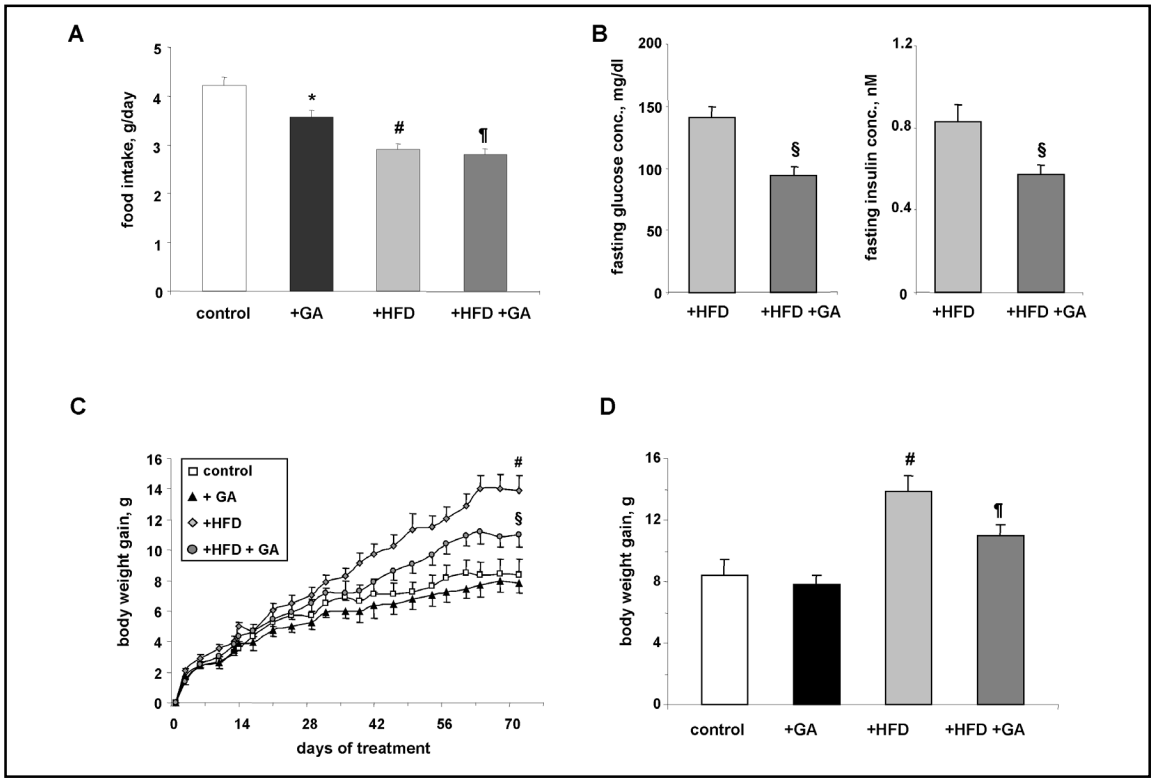


Fig. 5. Effect of GA treatment on fasting blood glucose, plasma insulin concentrations and obesity during high fat diet. A Arithmetic means ± SEM of mean daily food intake in untreated mice (n = 4) as well as mice treated with either GA (n=4) or high fat diet (HFD, n = 8) or both HFD and GA (n = 8) for 10 weeks. B: Arithmetic means ± SEM of fasting blood glucose and plasma insulin concentrations in mice treated with HFD or HFD + GA. C: Arithmetic means ± SEM of the body weight gain during 10 weeks in untreated mice as well as in mice treated with either GA (n = 4) or high fat diet (HFD, n = 8) or both HFD and GA (n = 8). D Arithmetic means ± SEM of total body weight gain within 10 weeks in untreated mice as well as in mice treated with either GA, with HFD or with both HFD and GA. *indicates significant difference between control and GA treatment #indicates significant difference between control and HFD treatment, ¶indicates significant difference between GA and HFD+GA treatment.

To test, whether the decreased intestinal SGLT1 activity influenced plasma glucose and insulin levels as well as body weight during excessive glucose ingestion, mice were treated for one week with tap water, 10% GA or a 20% (w/v) glucose solution with or without 10% GA. Prior to treatment with glucose, the plasma glucose concentration was not significantly different between GA-treated mice and control mice (Fig. 3A). Addition of 20% glucose to the drinking water significantly increased blood glucose concentrations, an effect significantly blunted by additional GA treatment. Excessive glucose intake tended to increase plasma insulin levels, an effect, however, not reaching statistical significance. The plasma insulin levels following excessive glucose intake tended to be lower in GA-treated animals, an effect, however, again not statistically significant (Fig. 3B).

Further experiments were performed to investigate the course of body weight under high glucose intake with or without GA for 4 weeks. Addition of glucose significantly increased fluid intake and decreased food intake to a similar extent in animals treated without or with GA (Fig. 4A, 4B). As shown in Figure 4C&D, administration of GA significantly blunted the body weight gain of mice drinking 20% glucose solution.

To test, whether GA is similarly effective in high fat diet, the body weight was monitored in animals receiving a high fat diet with or without GA. The treatment with GA did not significantly affect daily fluid intake (data not shown) and was followed by a slight, but significant decrease of food intake (Fig. 5A). As illustrated in Figure 5B, high fat diet was paralleled by hyperglycemia and hyperinsulinism, effects significantly blunted by GA treatment. Moreover, the treatment was followed by body weight gain, which was significantly blunted following GA treatment (Fig. 5 C&D).

Discussion

The present study reveals that GA decreases the membrane abundance of SGLT1, the major carrier accomplishing intestinal glucose absorption [10]. Accordingly, GA treatment decreases electrogenic glucose transport in Ussing chamber experiments. It is noteworthy that the decline of electrogenic glucose transport following GA treatment did not require the presence of GA in the experimental solutions during the Ussing chamber experiments and that the inhibitory effect of GA on electrogenic intestinal glucose transport thus cannot be attributed to a direct interaction of GA with the carrier but results from the decreased SGLT1 protein abundance in the brush border membrane. According to the gene array data, the decreased SGLT1 protein expression is not the result of decreased SGLT1 transcription but may rather be due to posttranscriptional regulation, such as interference with carrier trafficking or protein stability. Previous studies have revealed that SGLT1 may be subject to regulation of transcription [15], mRNA stability [16], transporter protein abundance in the plasma membrane [17], and transporter activity [18]. Factors modifying SGLT1 activity include carbohydrate-rich diet [19], adrenergic innervation [20], insulin [21], glucagon-like peptide 2 [22], cholecystokinin [17], insulin-like growth factors [23], cytosolic Na^+ [24] and lipopolysaccharides [25]. Moreover, SGLT1 activity is regulated by phosphatidylinositol (PI) 3 kinase [26], the phosphoinositide-dependent kinase 1 (PDK1) [27] as well as the serum-and glucocorticoid-regulated kinase isoforms SGK1 and SGK3 [14, 28], kinases regulating the transport of a variety of nutrients and channels [14, 28-33]. SGLT1 activity is further subject to downregulation by the 67-kDa-protein RS1 [11]. According to gene array analysis, decreased SGLT1 protein abundance could have been secondary to enhanced expression of RS1 (encoded by rsc1a1) or secondary to decreased expression of SGK3. Animals deficient of SGK3 indeed display decreased intestinal electrogenic glucose transport [14]. The present observations do not allow safe conclusions regarding the mechanisms underlying the decreased SGLT1 protein abundance in the intestinal brush border membrane following GA treatment.

Previous studies did not yield evidence for a direct inhibition of intestinal glucose uptake by GA [26]. In those experiments 2.5 g/l GA was applied directly to perfused jejunal segments. In our study, addition of GA (100 g/l) to the perfusate similarly did not inhibit the glucose-induced short-circuit currents. This observation again excludes the possibility that the effects of GA are due to direct inhibition of the carrier or due to chemical interaction and binding of glucose.

The downregulation of SGLT1 protein expression and activity were expected to influence plasma and insulin glucose concentrations. Indeed, the hyperglycemic effect of excessive glucose intake was significantly blunted by GA treatment. Moreover, GA treatment prevented the body weight gain following a four week treatment with glucose-rich diet despite similar fluid and food intake in untreated and GA-treated mice. A diet rich in dietary fibres is associated with reduced body weight [27] and prevention of metabolic syndrome [28]. In patients with diabetes mellitus type 2, an increased intake of dietary fibers improved glycemic control and reduced hyper-insulinemia [29]. Proposed explanations for the beneficial effects of dietary fibers include interaction with food intake and body weight through satiety, glycemia and insulinemia, blood lipids and blood pressure [30]. Interactions with intestinal glucose uptake have hitherto not been explored. In view of the present observations, other dietary fibers may be similarly effective at least in part by decreasing SGLT1 activity.

In summary, the dietary fiber GA inhibits intestinal glucose absorption via interaction with the membrane abundance of SGLT1. The effect could serve to prevent obesity and the development of metabolic syndrome.

Acknowledgements

The study was supported by the Deutsche Forschungsgemeinschaft. O. N. was supported by a research fellowship of the DAAD.

References

1 Younes H, Garleb K, Behr S, Remesy C, Demigne C: Fermentable fibers or oligosaccharides reduce urinary nitrogen excretion by increasing urea disposal in the rat cecum. J Nutr 1995;125:1010-1016.

2 W. D. Deckwer, B. Dill, E. Eisenbrand, U. Bornscheuer, A. Pühler, R. R. Heiker, A. Kirschnig, P. Schreier, P. Schreier, B. Fugmann, G. Pohnert, T. Gamse and H. Hulpke, Römpp, online, Georg-Thieme-Verlag, Stuttgart 2006.

3 Tiss A, Carriere F, Verger R: Effects of gum arabic on lipase interfacial binding and activity. Anal Biochem 2001;294:36-43.

4 Phillips GO: Acacia gum (Gum Arabic): a nutritional fibre; metabolism and calorific value. Food Addit Contam 1998;15:251-264.
5 Anderson DM: Evidence for the safety of gum arabic (Acacia senegal (L.) Willd.) as a food additive—a brief review. Food Addit Contam 1986;3:225-230.
6 Nasir O, Artunc F, Saeed A, Kambal MA, Kalbacher H, Sandulache D, Boini KM, Jahovic N, Lang F: Effects of gum arabic (Acacia senegal) on water and electrolyte balance in healthy mice. J Ren Nutr 2008;18:230-238.
7 Ibrahim MA, Kohn N, Wapnir RA: Proabsorptive effect of gum arabic in isotonic solutions orally administered to rats: effect on zinc and other solutes. J Nutr Biochem 2004;15:185-189.
8 Teichberg S, Wingertzahn MA, Moyse J, Wapnir RA: Effect of gum arabic in an oral rehydration solution on recovery from diarrhea in rats. J Pediatr Gastroenterol Nutr 1999;29:411-417.
9 Wapnir RA, Wingertzahn MA, Moyse J, Teichberg S: Gum arabic promotes rat jejunal sodium and water absorption from oral rehydration solutions in two models of diarrhea. Gastroenterology 1997;112:1979-1985.
10 Wright EM, Martin MG, Turk E: Intestinal absorption in health and disease—sugars. Best Pract Res Clin Gastroenterol 2003;17:943-956.
11 Osswald C, Baumgarten K, Stumpel F, Gorboulev V, Akimjanova M, Knobeloch KP, Horak I, Kluge R, Joost HG, Koepsell H: Mice without the regulator gene Rsc1A1 exhibit increased Na+-D-glucose cotransport in small intestine and develop obesity. Mol Cell Biol 2005;25:78-87.
12 Dieter M, Palmada M, Rajamanickam J, Aydin A, Busjahn A, Boehmer C, Luft FC, Lang F: Regulation of glucose transporter SGLT1 by ubiquitin ligase Nedd4-2 and kinases SGK1, SGK3, and PKB. Obes Res 2004;12:862-870.
13 Biber J, Stieger B, Haase W, Murer H: A high yield preparation for rat kidney brush border membranes. Different behaviour of lysosomal markers. Biochim Biophys Acta 1981;647:169-176.
14 Lang F, Busch GL, Ritter M, Volkl H, Waldegger S, Gulbins E, Haussinger D: Functional significance of cell volume regulatory mechanisms. Physiol Rev 1998;78:247-306.
15 Martin MG, Wang J, Solorzano-Vargas RS, Lam JT, Turk E, Wright EM: Regulation of the human Na(+)-glucose cotransporter gene, SGLT1, by HNF-1 and Sp1. Am J Physiol Gastrointest Liver Physiol 2000;278:G591-G603.
16 Loflin P, Lever JE: HuR binds a cyclic nucleotide-dependent, stabilizing domain in the 3' untranslated region of Na(+)/glucose cotransporter (SGLT1) mRNA. FEBS Lett 2001;509:267-271.
17 Hirsh AJ, Cheeseman CI: Cholecystokinin decreases intestinal hexose absorption by a parallel reduction in SGLT1 abundance in the brush-border membrane. J Biol Chem 1998;273:14545-14549.
18 Vayro S, Silverman M: PKC regulates turnover rate of rabbit intestinal Na+-glucose transporter expressed in COS-7 cells. Am J Physiol 1999;276:C1053-C1060.
19 Ferraris RP, Diamond JM: Specific regulation of intestinal nutrient transporters by their dietary substrates. Annu Rev Physiol 1989;51:125-141.
20 Ishikawa Y, Eguchi T, Ishida H: Mechanism of beta-adrenergic agonist-induced transmural transport of glucose in rat small intestine. Regulation of phosphorylation of SGLT1 controls the function. Biochim Biophys Acta 1997;1357:306-318.
21 Stumpel F, Kucera T, Gardemann A, Jungermann K: Acute increase by portal insulin in intestinal glucose absorption via hepatoenteral nerves in the rat. Gastroenterology 1996;110:1863-1869.
22 Cheeseman CI: Upregulation of SGLT-1 transport activity in rat jejunum induced by GLP-2 infusion in vivo. Am J Physiol 1997;273:R1965-R1971.
23 Lane RH, Dvorak B, MacLennan NK, Dvorakova K, Halpern MD, Pham TD, Philipps AF: IGF alters jejunal glucose transporter expression and serum glucose levels in immature rats. Am J Physiol Regul Integr Comp Physiol 2002;283:R1450-R1460.
24 Kusche-Vihrog K, Segal A, Grygorczyk R, Bangel-Ruland N, Van Driessche W, Weber WM: Expression of ENaC and other transport proteins in Xenopus oocytes is modulated by intracellular Na+. Cell Physiol Biochem 2009;23:9-24.
25 Amador P, Marca MC, Garcia-Herrera J, Lostao MP, Guillen N, de la OJ, Rodriguez-Yoldi MJ: Lipopolysaccharide induces inhibition of galactose intestinal transport in rabbits in vitro. Cell Physiol Biochem 2008;22:715-724.
26 Rexhepaj R, Artunc F, Metzger M, Skutella T, Lang F: PI3-kinase-dependent electrogenic intestinal transport of glucose and amino acids. Pflugers Arch 2007;453:863-870.
27 Artunc F, Rexhepaj R, Volkl H, Grahammer F, Remy C, Sandulache D, Nasir O, Wagner CA, Alessi DR, Lang F: Impaired intestinal and renal glucose transport in PDK-1 hypomorphic mice. Am J Physiol Regul Integr Comp Physiol 2006;291:R1533-R1538.
28 Schwab M, Lupescu A, Mota M, Mota E, Frey A, Simon P, Mertens PR, Floege J, Luft F, Asante-Poku S, Schaeffeler E, Lang F: Association of SGK1 gene polymorphisms with type 2 diabetes. Cell Physiol Biochem 2008;21:151-160.
29 Boehmer C, Palmada M, Klaus F, Jeyaraj S, Lindner R, Laufer J, Daniel H, Lang F: The peptide transporter PEPT2 is targeted by the protein kinase SGK1 and the scaffold protein NHERF2. Cell Physiol Biochem 2008;22:705-714.
30 Boehmer C, Laufer J, Jeyaraj S, Klaus F, Lindner R, Lang F, Palmada M: Modulation of the voltage-gated potassium channel Kv1.5 by the SGK1 protein kinase involves inhibition of channel ubiquitination. Cell Physiol Biochem 2008;22:591-600.
31 Gehring EM, Zurn A, Klaus F, Laufer J, Sopjani M, Lindner R, Strutz-Seebohm N, Tavare JM, Boehmer C, Palmada M, Lang UE, Seebohm G, Lang F: Regulation of the glutamate transporter EAAT2 by PIKfyve. Cell Physiol Biochem 2009;24:361-368.
32 Krueger B, Haerteis S, Yang L, Hartner A, Rauh R, Korbmacher C, Diakov A: Cholesterol depletion of the plasma membrane prevents activation of the epithelial sodium channel (ENaC) by SGK1. Cell Physiol Biochem 2009;24:605-618.
33 Laufer J, Boehmer C, Jeyaraj S, Knuwer M, Klaus F, Lindner R, Palmada M, Lang F: The C-terminal PDZ-binding motif in the Kv1.5 potassium channel governs its modulation by the Na+/H+ exchanger regulatory factor 2. Cell Physiol Biochem 2009;23:25-36.
34 Wingertzahn MA, Teichberg S, Wapnir RA: Stimulation of non-sodium-dependent water, electrolyte, and glucose transport in rat small intestine by gum arabic. Dig Dis Sci 2001;46:1105-1112.
35 Koh-Banerjee P, Franz M, Sampson L, Liu S, Jacobs DR, Jr., Spiegelman D, Willett W, Rimm E: Changes in wholegrain, bran, and cereal fiber consumption in relation to 8-y weight gain among men. Am J Clin Nutr 2004;80:1237-1245.
36 McKeown NM, Meigs JB, Liu S, Saltzman E, Wilson PW, Jacques PF: Carbohydrate nutrition, insulin resistance, and the prevalence of the metabolic syndrome in the Framingham Offspring Cohort. Diabetes Care 2004;27:538-546.
37 Chandalia M, Garg A, Lutjohann D, von Bergmann K, Grundy SM, Brinkley LJ: Beneficial effects of high dietary fiber intake in patients with type 2 diabetes mellitus. N Engl J Med 2000;342:1392-1398.
38 Delzenne NM, Cani PD: A place for dietary fibre in the management of the metabolic syndrome. Curr Opin Clin Nutr Metab Care 2005;8:636-640.